

## Description

# METHOD TO TREAT PATIENTS WITH AMYOTROPHIC LATERAL SCLEROSIS AND THE LIKE

### BACKGROUND OF INVENTION

#### FIELD OF THE INVENTION:

[0001] This invention relates generally to the treatment of central nervous system (CNS) and peripheral nervous system (PNS) neuropathies, especially amyotrophic lateral sclerosis (ALS), wherein the neurotoxicity associated with the porphyrin precursors, delta-aminolevulinic acid (ALA) and porphobilinogen (PBG), is a contributing factor, and more particularly, to the use of a uroporphyrin isomer, uroporphyrin I (URO I) or uroporphyrin III (URO III) or any of their substrates as such treatment.

[0002] Normal hepatic heme synthesis is necessary to life in mammals. It is a multistage process (Fig. 1) that includes the porphyrin precursors, ALA (Fig.4) and PBG (Fig. 5) as

well as enzymes/catalysts and porphyrinogens, which are the progenitors of their respective porphyrins. The heme produced by the liver is incorporated into the hemoproteins that the liver uses to neutralize all toxins, whether they are of endogenous or exogenous origin. Levels of all of the components of hepatic heme synthesis are elevated when demand for hepatic hemoproteins to neutralize toxins is high except for the isomer I porphyrinogens, uroporphyrinogen I (UROGEN I) and coproporphyrinogen I (COPROGEN I), as they are not substrates of heme. Heme production is partially regulated by levels of production of the enzymes/catalysts, partially by the availability of iron and partially by feedback inhibition.

#### **HEPATIC PORPHYRIAS AND A RELATED DISORDER:**

[0003] The hepatic porphyrias are usually caused by inborn shortages of one or more of the enzymes/catalysts that facilitate heme synthesis (Nordmann Y; Puy H. 2002, Clin Chim Acta, 325(1-2):17-37), though some porphyrias, notably porphyria cutanea tarda (PCT) and porphyria turcica (Dean G. 1981, Arch Dermatol, (6):318), may be acquired.

[0004] The acute (neurotoxic) porphyrias cause CNS and PNS damage, elevations of porphyrins and may cause demen-

tia. Most cause cutaneous photosensitivity as well. (Lip GY. et al. 1993, Br J Clin Pract 47(1):38–43).

[0005] Though the precise pathogenesis of the neuron damage is unknown, elevations of the porphyrin precursors, ALA and/or PBG, are associated with the CNS and PNS destruction in several hepatic porphyrias, hereditary coproporphyria (HCP) being just one example (Kuhnel A. et al. 2000, Clin Biochem Aug;33(6):465–73), but most notably acute intermittent porphyria (AIP), which is the porphyria that produces the most neurological damage and exhibits the lowest production of URO I (Hindmarsh JT et al. 1999, Clin Biochem, 32(8):609–19). There are no porphyrin elevations in AIP and there is therefore no cutaneous photosensitivity. Since the enzyme/catalyst deficiency is *phobilinogen deaminase* (Lundin G et al. 1997, Hum Genet 100(1):63–6) and more rarely, *hydroxymethylbilane synthase* (Ong PM et al. 1998, Hum Hered 48(1):24–9) there is a buildup of the two precursors, ALA and PBG, before sufficient antidotal levels of the two uroporphyrin isomers can be produced. When supplies of heme are adequate, production of the porphyrin precursors slows. This feedback inhibition mechanism is the reason that hematin is the current treatment of choice to suppress

precursor production during acute attacks. Unfortunately, prophylactic use of hematin is not possible as it causes circulatory collapse (Khanderia U. 1986, Clin Pharm 5(8):690–2).

[0006] The only hepatic porphyria that is never acute (neurotoxic) is PCT which is characterized by excessive production of URO I. (Bygum A et al. 2003 Acta Derm Venereol 283(2):115–20) and also cutaneous photosensitivity. It is exacerbated by increases in iron levels and is treated by regularly scheduled phlebotomy, which reduces serum levels of iron. Menstruation makes it less severe in females of childbearing age. (Nishioka E et al. 2003, J Am Acad Dermatol, 49(3):547–50).

[0007] Another disease, currently known as hereditary biochemical multiple sclerosis (HBMS) (Rooney R.N. et al. 1999, Am J Med Genet, 86(2), 194–196) also causes motor neuron destruction indistinguishable from that seen in ALS except that neurons of the sensory cortex are also affected in HBMS. HBMS, which causes no PNS damage like the acute porphyrias do, is characterized by a shortage of URO I and an elevation in URO III (Fig. 6), indicating that URO I protects the CNS from precursor associated damage while URO III protects the PNS.

[0008] In HBMS, the shortage of URO I is caused by a lack of the iron that is needed to bridge levels of the enzymes/catalysts needed to support normal hepatic heme synthesis, uroporphyrinogen III synthase and cosynthase and uroporphyrinogen decarboxylase (Fig 6) (Ponka P.1997, Blood. 89, 1–25). In HBMS, daily ingestion of at least 350 mg. of iron reroutes the path of heme synthesis such that normal amounts of UROGEN I, and hence its product, URO I, are produced in a timely fashion (Rooney R.N. et al. unpublished data), which halts the disease progression.

[0009] In normal hepatic heme synthesis, porphyrin precursors are always produced; hence the associated neurotoxicity is also always present. URO I, the molecule that is the antidote to the neuron destruction associated with the porphyrin precursors, is also produced (Fig. 1). URO I cannot effectively counteract the neurotoxicity associated with the porphyrin precursors in the CNS unless it can enter into and circulate within the CNS. If it cannot, the CNS will sustain further damage in almost every instance of hepatic heme synthesis just as it does in the acute porphyrias.

[0010] Guamanian ALS has causes and effects (Banack SA et al. 2003 Neurology. 12;61(3), :387–9) (Ikemoto A et al 2000, Amyotroph Lateral Scler Other Motor Neuron Disord

. Mar;1(2):97–104) similar to those of the acquired acute porphyria, porphyria turcica (Cripps DJ. et al 1984, Br J Dermatol. 111(4), :413–22) that they may actually be the same disease though the toxic precipitator of Guamanian ALS, cycad nuts, was probably even more pernicious than the fungicides that precipitated porphyria turcica. Both were caused by ingested toxins that had contaminated the food supply and both, therefore, greatly increased demand for hepatic heme synthesis. Guamanian ALS was defined in 1952 (Donald Koerner and Jose Torres. 1952, Annals of Internal Medicine) and predated the Hoesch or Watson–Schwarz tests which quantify the porphyrin precursor, porphobilinogen (PBG), and are the still current and best tests for the acute neurotoxic porphyrias (Mauzerall D, Granick S., J Biol Chem 1956;219:435–446). Guamanian amyotrophic lateral sclerosis, therefore, could not have been accurately diagnosed as the acquired acute porphyria that it probably actually was.

[0011] The damage done by untreated HBMS is limited to damage to the motor cortex and to the sensory cortex. It is caused by a lack of the iron needed to support the production of normal amounts of URO I. ALS is most likely the result of the CNS becoming at least partially impermeable to URO I

or to an inability to distribute URO I as normal within the CNS. Otherwise, the effects on the CNS are very closely matched though the etiology and parts of the pathogenesis are different. Although HBMS is inherited via a pattern of autosomal dominance with variable penetrance and is not sex linked, males are less likely to suffer neurological damage because they do not have the monthly iron loss associated with menstruation.

#### **RELATIONSHIP TO AMYOTROPHIC LATERAL SCLEROSIS:**

- [0012] The rates of CNS destruction between the two "siblings", ALS and HBMS, should be different because there are times when even untreated HBMS sufferers are able to produce sufficient URO I due to normal fluctuations in iron intake and availability. During those times, production and delivery of URO I is normal and no further CNS damage is incurred.
- [0013] The second difference is that ALS does not include sensory deficit while HBMS does. Though future research will be required to precisely define this difference, it can be explained as follows: Untreated HBMS includes periods when production of the neurotoxicity associated porphyrin precursors is high while URO I production is low. CNS damage is inevitable during those periods.

[0014] In ALS, except for Guamanian ALS, there is no disturbance in hepatic heme synthesis so normal amounts of the porphyrin precursors are produced as is URO I and all enter the bloodstream in a time interval that is effective for preserving the CNS.

[0015] A plausible explanation of why ALS affects only the motor neurons while HBMS affects both sensory and motor neurons is that the ALS brain may remain partially permeable to URO I or mutated superoxide dismutase-1 (SOD1) structures may incompletely absorb URO I, allowing enough URO I into the CNS to protect the sensory cortex but not enough to continue on and protect the motor cortex as well.

[0016] This concept is supported by the very existence of Guamanian ALS because it is an example of the neurotoxicity associated porphyrin precursor production outpacing URO I production. That imbalance would explain why the effects of Guamanian ALS on the CNS would be identical to that of familial amyotrophic lateral sclerosis (FALS) and sporadic ALS even though their etiologies and probably their pathogeneses are different. URO I delivery would be timely but amounts would be insufficient. This echoes the pattern of the CNS damage of AIP.



[0017] URO I, if normal, has a small enough molecular size (Fig. 2) to penetrate the normal BBB while its isomer, URO III (Fig. 3), which is the product of uroporphyrinogen III, which is on the direct path to heme, is a larger molecule that cannot.

[0018] There is no record of a deficit of URO I or a surfeit of the porphyrin precursors in the serum or urine of ALS patients though that was necessarily true for Guamanian ALS and evidently went undetected, unrecorded or was ignored. There has also been no evidence that the URO I molecule produced in all cases of ALS has the normal structure (Fig. 2) that is best suited to penetrating the blood-brain barrier. There has been evidence of structures in the ALS brain that evidently either make the BBB less permeable to URO I or prevent its circulation to the motor neurons. The neurotoxicity associated with the porphyrin precursors is not blocked by the BBB, as is evidenced by the acute porphyrias. This may be due to the fact that a single molecule of porphobilinogen is even smaller than either of the two uroporphyrin isomers. (Fig. 5 as compared to Fig. 2 and Fig. 3).

[0019] SOD1 (Kato, S. et al. 2001, Neuropathology. 21(1): 67–81) found in familial ALS (FALS), is likely to be a cause of the

blockage of the entry of URO I into the CNS or of inhibiting its distribution to the motor neurons by the neuronal Lewy body-like hyaline inclusions (LBHI) and astrocytic hyaline inclusions (Ast-HI) that are the hallmarks of FALS. The logic is that these products of the mutated SOD1 either clog the BBB such that entry of URO I into the CNS is either inhibited or halted altogether or that the structures produced by the SOD1 mutation absorb URO I preventing it from circulating as normal within the brain.

[0020] PCT, the most common of the porphyrias, is the only hepatic porphyria that is never neurotoxic and is the only hepatic porphyria characterized by excessive production of URO I. It is not known if Stephen Hawking, the most prominent example of a patient with slowly progressing ALS, has concomitant ALS and PCT.

[0021] There has been at least one report of an acute porphyria concomitant with PCT in which no nervous system damage was reported. (Doss MO et al. 2002, Med Klin (Munich) 97(1):1–5).

[0022] There has also been a report of an iron porphyrin (FeTCPP) being helpful in slowing disease progression in a mouse of ALSAS et al. 2003,. J Neurochem 85(1):142–50) This may be related to the feedback inhibition of the neurotoxicity

associated porphyrin precursors, ALA and porphobilinogen (PBG) and may therefore not be safely sustainable on a long term basis just as hematin is not.

## **OTHER DISORDERS CAPABLE OF STIMULATING HEPATIC HEME**

### **SYNTHESIS:**

[0023] Most illnesses, injuries, infections, disorders or toxins of endogenous or exogenous origin stimulate hepatic heme synthesis, meaning that any that also involve the PNS and/or the CNS can be exacerbated by any of those triggers unless they are accompanied by commensurate increases in either or both of the uroporphyrin isomers, uroporphyrins I and/or III, meaning that CNS disorders that can be ameliorated by URO I include, but are not limited to, ALS, stroke, encephalitis, meningitis, and hereditary biochemical multiple sclerosis (HBMS). PNS disorders that can be ameliorated by URO III include, but are not limited to, AIDS related neuropathy, Guillaine-Barre syndrome and diabetic neuropathy.

## **SUMMARY OF INVENTION**

[0024] The present invention teaches a hitherto unknown value, purpose and use for naturally produced molecules that had hitherto been considered valueless. They are neuro-protectors, which can be used to halt, mitigate, prevent or

preclude the neurological damage associated with other naturally produced molecules, the porphyrin precursors, ALA and PBG, that are also part of the hepatic heme biosynthetic pathway. The neuroprotectors are useful in halting or mitigating the neuron damage in some common neurological diseases, disorders and injuries. This invention gives rise to the objectives described below.

[0025] An objective of the invention is to use URO I to halt or mitigate the CNS damage that would otherwise occur as a result of the neurotoxicity associated with the presence of the porphyrin precursors, ALA and/or PBG.

[0026] Another objective of the invention is to use URO III to halt or mitigate the PNS damage that would otherwise occur as a result of the neurotoxicity associated with the presence of the porphyrin precursors, ALA and/or PBG.

#### **BRIEF DESCRIPTION OF DRAWINGS**

[0027] The accompanying drawings illustrate the present invention. In these drawings:.

[0028] Fig. 1 is a flow diagram that illustrates the position in heme synthesis normally occupied by UROGEN I.

[0029] Fig. 2 is a diagram of the molecular structure of URO I.

[0030] Fig. 3 is a diagram of the molecular structure of URO III.

[0031] Fig. 4 is a diagram of the molecular structure of ALA.

[0032] Fig. 5 is a diagram of the molecular structure of PBG.

[0033] Fig. 6 is a flow diagram of heme synthesis in untreated HBMS.

#### **DETAILED DESCRIPTION**

[0034] Referring now to Figure 1, there is shown a flow diagram of normal heme synthesis. The heme biosynthetic pathway is a multistage process starting with glycine and succinyl-CoA and facilitated by several enzymes/catalysts that result in the end product of heme. The part of heme synthesis relevant to this invention begins with aminolevulinic acid synthase and culminates with heme. There is a straight line progression from aminolevulinic acid synthase to heme except for a spontaneous conversion of hydroxymethylbilane (HMB) to UROGEN I, progenitor of URO I, when uroporphyrinogen III (UROGEN III) synthase and UROGEN III co-synthase, which are steps in the path to heme and the progenitors of URO III, do not convert all of the available HMB to uroporphyrinogen III (UROGEN III). (Sassa S. 1996, Blood Review, 10, 53–58).

[0035] Referring now to Figure 2, there is shown a diagram of the molecular structure of URO I. Normal URO I is the smaller

of the two uroporphyrin isomer molecules and is therefore the one more capable of penetrating the BBB.

[0036] Referring now to Figure 3, there is shown a diagram of the molecular structure of URO III. URO III is the larger of the two uroporphyrin isomer molecules and is therefore the one that is less capable of penetrating the BBB.

[0037] Referring now to Figure 4, there is shown a diagram of the molecular structure of ALA. Aminolevulinic acid is one of the two porphyrin precursors that has been associated with neuron damage.

[0038] Referring now to Figure 5, there is shown a diagram of the molecular structure of porphobilinogen (PBG). Porphobilinogen is the other porphyrin precursor that has been associated with neuron damage.

[0039] Referring now to Figure 6, there is shown a diagram of the heme biosynthetic pathway in untreated HBMS. Comparisons to normal amounts are indicated by enlargement or reduction of type fonts as enzymes compare to enzymes, uroporphyrinogens (UROGENS) compare to UROGENS and coproporphyrinogens (COPROGENS) compare to COPROGENS. When there is insufficient iron availability in the liver, levels of uroporphyrinogen III synthase and cosynthase and of uroporphyrinogen decarboxylase are all

overproduced. Overproduction of those enzymes/catalysts causes reductions in UROGEN I production and thereby causes increases of both of the coproporphyrinogen isomers, further depleting what little UROGEN I had been produced. This illustration is based upon the typical porphyrin counts of urinalyses during acute attack in a patient who had HBMS. This porphyrin distribution is based on several porphyrin urinalyses measured during exacerbations. (Rooney RN et al. unpublished data) That patient's disease is now arrested as a result of daily iron supplementation. (Rooney RN, et al. 1999, Am J Med Genet, 86(2), 194–196).

[0040] The above described drawings illustrate the background information necessary to understand the unique and previously unrecognized value of uroporphyrin(ogen) I (URO I), its position in the flow of heme synthesis, and its comparative molecular size in relation to the other pertinent molecules involved in hepatic heme synthesis.

[0041] The background information details why URO I, whose importance as a neuroprotector had not previously been recognized, is actually vital to healthy CNS function. Since that value has now been recognized thanks to the discovery of HBMS, it must be assured that URO I be made avail-

able to the CNS whenever the porphyrin precursors and their associated neurotoxicity are present.

[0042] URO I is currently readily available on the medical marketplace for use in laboratory experiments and has been used for years to piggyback antibodies that attach to cancer cells so that the photosensitive properties of porphyrins would then render the cancer cells vulnerable to destruction by light.

[0043] Implementation of this invention contemplates a medication that entails the purification of URO I and URO III to medical grade standards, determination of formulations suitable for the various possible modes of delivery, determination of correct dosages and Food and Drug Administration (FDA) approval for its use as a medication for humans.

[0044] This invention was made possible by the discovery of HBMS, which supplies the missing link that connects and gives new meaning to research done more than fifty years ago and on through research completed in 2003, less than fifty weeks ago.

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#### **PARAMETERS:**

- [0062] Since other modifications and changes varied to fit particular operating requirements and environments will be apparent to those skilled in the art, the invention is not limited to the examples chosen for purposes of disclosure, and covers all changes and modifications which do not

constitute departures from the true spirit and scope of this invention.

[0063] Having thus described the invention, what is desired to be protected by Letters Patent is presented in the subsequently presented claims.